THE PHOTOSENSITIVE RETINAL PIGMENTS OF FISHES FROM RELATIVELY TURBID COASTAL WATERS

By FREDERICK W. MUNZ*

(From the Department of Zoology, University of California, Los Angeles)

(Received for publication, April 24, 1958)

ABSTRACT

Digitonin extracts have been prepared from the retinae of a dozen species of marine and euryhaline teleost fishes from turbid water habitats. Spectrophotometric analysis of the extracts shows that the photosensitive retinal pigments of these species have maximum absorption above 500 mμ. In nine species there are retinene$_1$ pigments with λ_{max} between 504 and 512 mμ. In the marine but euryhaline mullet, *Mugil cephalus*, there is a porphyropsin with λ_{max} 520 mμ. A mixture of rhodopsin and porphyropsin in an extract of a marine puffer, *Sphoeroides annulatus*, was disclosed by partial bleaching with colored light. In addition, one other species has a 508 mμ pigment, of which the nature of the chromophore was not determined.

The habitats in which these fishes live are relatively turbid, with the water greenish or yellowish in color. The spectral transmission of such waters is probably maximal between 520 and 570 mμ. It is suggested that the fishes have become adapted to these conditions by small but significant shifts in spectral absorption of their retinal pigments. These pigments are decidedly more effective than rhodopsin in absorption of wavelengths above 500 mμ. This offers a possible interpretation of the confusing array of retinal pigments described from marine and euryhaline fishes.

In the older view of visual pigment distribution in the vertebrates (Wald, 1953) there was an elegant simplicity in the nature and arrangement of the chemical constituents. In particular, proteins in the rods were believed to combine with retinene$_1$ in terrestrial vertebrates and marine fishes to form rhodopsins (λ_{max} 500 ± 2 mμ). These rod proteins were thought to be very similar in different species; in fresh water fishes they were believed to combine with retinene$_2$ to produce porphyropsins (λ_{max} 522 ± 2 mμ). Recent experiments have shown that the "rhodopsins" of different species have λ_{max} ranging at least from 478 mμ (Munz, 1958) to 524 mμ (Crescitelli, 1956). "Porphyropsins" also are not limited to a single spectral position (*e.g.*, Dartnall, 1952). Morton and Pitt (1957) have published a review of the more recent work on visual pigments, but new pigments are being described at an increasingly rapid rate (*e.g.*, Wald, Brown, and Brown, 1957).

* This work is a portion of a dissertation for the Ph.D. degree at the University of California, Los Angeles.

In the spectrophotometric studies of visual pigments there have been few attempts to interpret the findings in terms of their possible physiological or ecological significance. Willmer (*in* Jones and Eckstein, 1956, p. 101) suggested that the distribution of rhodopsin and porphyropsin may be determined by the role of vitamin A in salt or water balance. This possibility that the visual pigments are determined by another, more primary process might adequately account for the occurrence of retinene$_1$ and retinene$_2$ photopigments, but would leave the large differences in spectral position within each group to be dismissed as chance variation.

With the discovery of a series of new light-sensitive pigments in deep sea fishes, Denton and Warren (1956), measuring the over-all changes in optical density following exposure of the retinae to white light, and Munz (1957), who analyzed retinal extracts, were led to suggest that the spectral locations of these pigments may be correlated with the predominant color of the light available to the fishes. It has even been proposed (Munz, 1958) that there may be a correlation with the dominant wavelengths of bioluminescence.

If it is true that the bathypelagic fishes, living in a blue environment, have visual pigments absorbing light maximally in the blue region of the spectrum, then perhaps fishes that characteristically live in greenish or yellowish waters might be expected to show a shift in their visual pigments toward longer wavelengths. The waters of ocean bays, river estuaries, and sandy beaches transmit light maximally of wavelengths between about 520 and 570 mμ (Clarke and James, 1939; Hulburt, 1945). The mudsucker, *Gillichthys mirabilis* Cooper, is a marine fish living in turbid water environments. The retinal pigment of this species is based on retinene$_1$, but has its λ_{max} 512 mμ (Munz, 1956), at a significantly longer wavelength than most other "rhodopsins."

This paper describes experiments which show that the retinal pigment of *Gillichthys* is not an isolated case, but is illustrative of a general trend in marine and euryhaline fishes from turbid coastal waters.

Materials and Methods

Eleven species of teleost fishes (Table I) were seined from sandy beaches, bays, or stream estuaries of the California coast in the period from 1955 to 1957. Unfortunately two species of the genus *Cottus* were not kept separate. These species are *aleuticus* Gilbert and *asper* Richardson. Two additional species seined in the Gulf of California, Mexico, were *Mugil cephalus* Linnaeus, taken at two locations not far from San Felipe, Baja California, and *Sphoeroides annulatus* (Jenyns) from near Guaymas, Sonora.

These fishes are not all limited to the same salinity, but occur in a variety of habitats. Strictly marine are *Anchoa compressa* (Girard), *Atherinopsis californiensis* Girard, *Seriphus politus* Ayres, *Embiotoca jacksoni* Agassiz, and *Hyperprosopon argenteum* Gibbons. The following species spawn in the sea and only occasionally enter fresh water: *Atherinops affinis* (Ayres), *Leptocottus armatus* Girard, *Clevelandia ios* (Jordan and Gilbert), and *Sphoeroides annulatus*. *Mugil cephalus* spawns in the sea

and often enters fresh water. *Eucyclogobius newberryi* (Girard) occurs in either fresh or brackish water, and *Cottus aleuticus* and *asper* are essentially fresh water fishes, occasionally going into brackish water.

TABLE I

Spectral Characteristics of Retinal Extracts

Species	Family	No. of eyes	Extract sample	Density 700 mμ	Maximum density	λ_{max}	Ratio	Adjusted λ_{max}
						mμ		mμ
*Anchoa compressa**	Engraulididae	14	1A‡	0.006	—§	—	—	—
*Atherinops affinis**	Atherinidae	36	2A	0.004	0.258	498	0.90	510
			2B	0.002	—§	—	—	—
*Atherinopsis californiensis**	Atherinidae	6	3A	0.029	0.790	488	0.97	—
Mugil cephalus	Mugilidae	10	4A	0.003	0.149	518	0.50	520
			4B	0.003	0.148	518	0.49	520
		10	5A	0.039	0.752	513	0.66	518
			5B	0.070	0.750	512	0.79	520
			5C	0.042	0.540	511	0.77	519
*Seriphus politus**	Sciaenidae	4	6A	0.007	0.847	500	0.61	503
			6B	0.003	0.477	496	0.83	504
*Embiotoca jacksoni**	Embiotocidae	6	7A	0.005	0.732	500	0.67	505‖
			7B	0.006	0.760	497	0.70	502‖
*Hyperprosopon argenteum**	Embiotocidae	2	8A	0.008	0.757	503	0.65	507‖
*Cottus sp.**	Cottidae	40	9A‡	0.025	0.595	504	0.76	511
*Leptocottus armatus**	Cottidae	38	10A‡	0.020	—§	—	—	—
		28	11A	0.007	0.942	500	0.55	503¶
			11B	0.008	0.777	499	0.60	502¶
			11C	0.006	0.292	494	0.63	498¶
*Clevelandia ios**	Gobiidae	36	12A‡	0.006	0.116	502	0.87	512
*Eucyclogobius newberryi**	Gobiidae	60	13A‡	0.004	—§	—	—	—
			13B‡	0.010	—§	—	—	—
		60	14A‡	0.016	0.305	502	0.87	512
Sphoeroides annulatus	Tetraodontidae	14	15A	0.002	0.150	495	0.89	508‖

* Taken in California.

‡ Whole eye extract.

§ Extract impure, ratio greater than 1.

‖ Mixture of two light-sensitive components.

¶ Another, light-stable pigment was present in the extract.

Digitonin extracts of the dark-adapted retinae of these fishes were prepared in dim red light with methods described elsewhere (Munz, 1956). The extracts were adjusted to pH 8.3 with borate-KCl buffer and were stored in darkness at 5–10°C. until analyzed.

After centrifugation the retinal extracts were analyzed in a Beckman DU spectrophotometer with photomultiplier attachment. Two per cent digitonin solution was

used as a blank. Optical density of the extracts (1 cm. light path) was measured from 700 to 340 mμ at 20 mμ intervals, followed by an interlaced return series of measurements, also at 20 mμ intervals. A Bausch and Lomb grating monochromator with interference filters placed in the exit light path provided narrow band colored light (half band widths between 10 and 15 mμ) used in bleaching the extracts. Following exposure to light the extracts were returned to the spectrophotometer and the absorption spectra measured as before. In this way the effects on each extract of several bleaching wavelengths were studied successively. Temperature was regulated at 20 ± 1°C. during measurements and bleaching. White light from a 60 watt tungsten bulb was also employed as a bleaching source.

RESULTS

1. Two Euryhaline Gobies

Both *Clevelandia ios* (arrow goby) and *Eucyclogobius newberryi* (tidewater goby), members of the family Gobiidae, have photosensitive retinal pigments very similar to that of another goby, *Gillichthys mirabilis*, also from coastal habitats. In all three species the light-sensitive pigment is based on retinene$_1$ and has λ_{max} 512 mμ. The observations described here are similar to those obtained with retinal extracts of *Gillichthys* (Munz, 1956). The absorption spectra of samples 12A of *Clevelandia* and 14A of *Eucyclogobius* had λ_{max} 502 mμ. Because of the small size of these fishes, the whole eyes had been crushed and treated with digitonin. This procedure gave rather impure extracts with a ratio of optical densities at the points of minimum and maximum absorption ("purity ratio") of 0.87 (Table I, ratio). Employing the same technique that Crescitelli and Dartnall (1954) used in an analysis of porphyropsin from the carp, the true λ_{max} was estimated to be 512 mμ (adjusted λ_{max}).

Partial bleaching experiments with narrow band colored light furnished confirmation of this estimate in both species. Sample 12A of *Clevelandia* was treated with hydroxylamine (0.1 ml. of 0.1 M NH_2OH was added to both sample and blank) and bleached in stages with red (640 mμ) and orange-red (606 mμ) light. Maximal density losses due to bleaching occurred at 512 and 511 mμ, respectively. Samples 13A and B of *Eucyclogobius* were also treated with NH_2OH and subjected to partial bleaching with 640, 606, and 390 mμ light and with white light. Only a single component was bleached, λ_{max} of the difference spectra being 512 to 514 mμ. The difference spectra obtained with these species are compared (Fig. 1) with that of the crested goby, *Coryphopterus nicholsii* (Bean), another member of the same family. *Coryphopterus*, however, typically lives along rocky coasts where the water is clear. It does not have the 512 mμ pigment, but has ordinary rhodopsin with λ_{max} 500 mμ.

In all these related species the product of bleaching is characteristic of retinene$_1$ pigments (Crescitelli, 1956). In NH_2OH experiments with *Clevelandia* and *Eucyclogobius* the product (*i.e.*, oxime) had λ_{max} 368 to 370 mμ, the spectral position of retinene$_1$ oxime. The photosensitive pigments of *Clevelandia* and

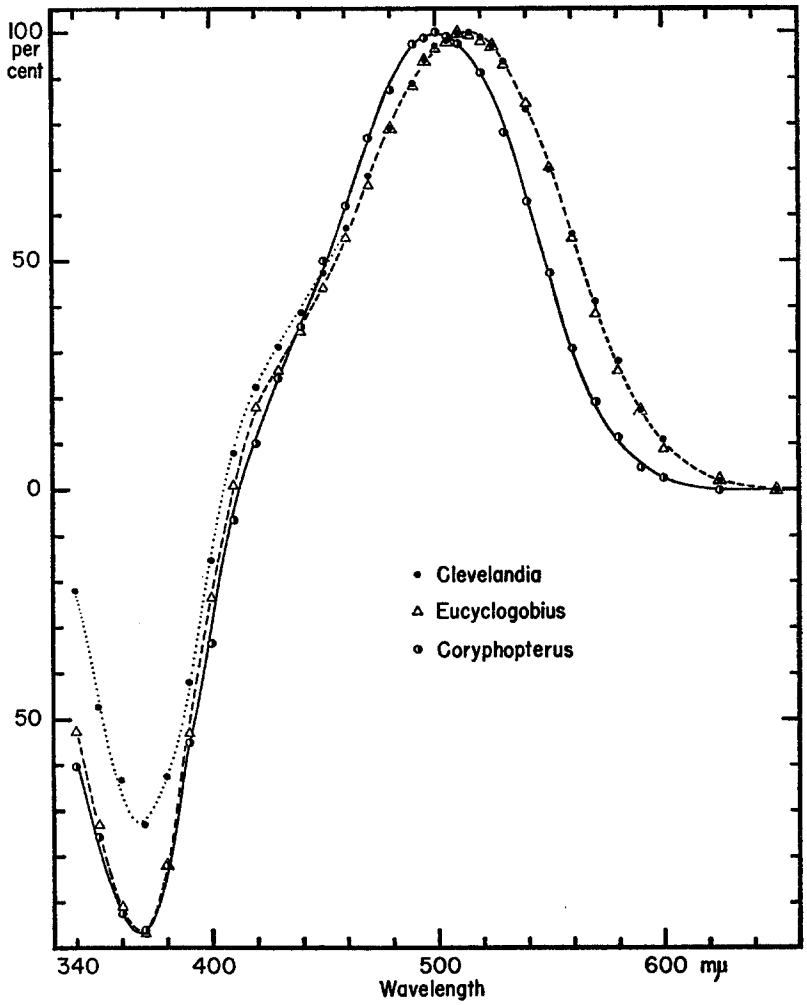

FIG. 1. Comparison of NH_2OH difference spectra of three gobies. All curves are scaled to 100 per cent for comparison. *Clevelandia ios*, sample 12A, combined 640 and 606 mμ bleaches; *Eucyclogobius newberryi*, sample 13A, combined 640 and 606 mμ bleaches; *Coryphopterus nicholsii*, 580 mμ bleach.

Eucyclogobius are 512_1 pigments (subscript indicates that retinene$_1$ is the chromophore) like that of *Gillichthys*.

2. *Euryhaline Cottid Fishes*

Two genera of euryhaline cottids (sculpins) had the same photosensitive pigment. Since the extract (sample 9A) of *Cottus*, the chiefly fresh water genus,

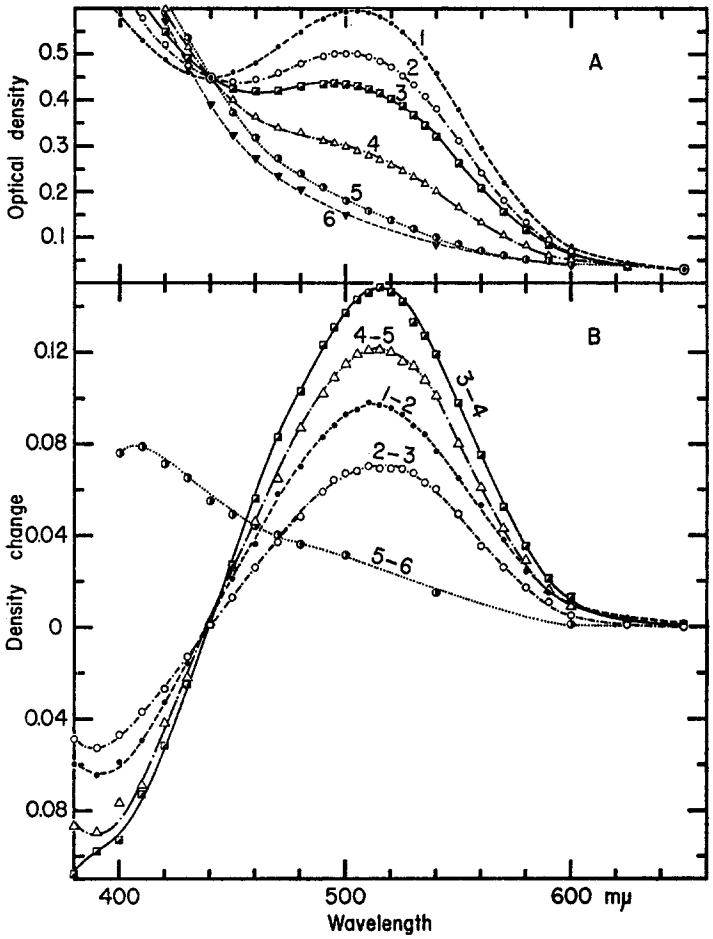

FIG. 2 A. A partial bleaching experiment with sample 9A of *Cottus sp.* Curve 1, absorption spectrum of unbleached extract; curve 2, after 1 hour exposure to 640 mμ light; curve 3, after 1 hour further exposure to 640 mμ light; curve 4, after 15 minutes' exposure to 606 mμ light; curve 5, after 43 minutes' further exposure to 606 mμ light; curve 6, after 10 minutes' exposure to white light.

FIG. 2 B. Difference spectra of experiment with sample 9A. Upward changes indicate loss of density, downward changes gain in density. Curves 1–2 and 2–3 are the results of the 640 mμ bleaches; 3–4 and 4–5, the 606 mμ bleaches; and 5–6, the white light bleach.

gave no evidence of a mixture of photosensitive pigments, it seems likely that both species probably included (*aleuticus* and *asper*) share the same or very similar pigments. The estimated true λ_{max} was 511 mμ (Table I). Partial bleaching revealed only a single photosensitive component (Fig. 2), λ_{max} of the difference spectrum being 513 mμ. In an experiment like this without NH_2OH the

maximum of the difference spectrum is shifted to a slightly longer wavelength by interaction with the product of bleaching (Crescitelli and Dartnall, 1954); the difference spectrum of *Cottus* therefore confirms the estimate of 511 mμ as λ_{max}. The product of bleaching (without NH_2OH) absorbed maximally at 386

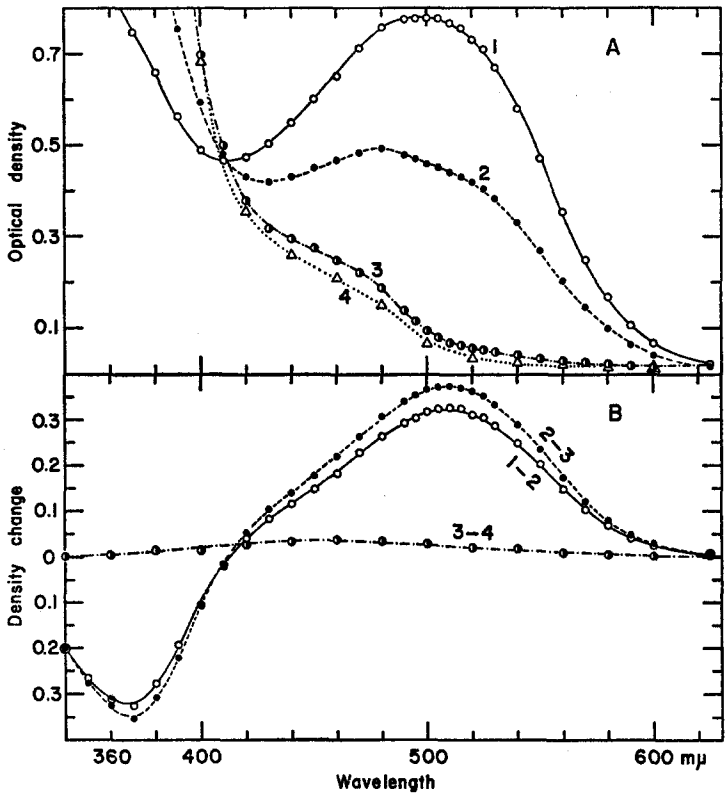

FIG. 3 A. A partial bleaching experiment with sample 11B of *Leptocottus armatus* (NH_2OH added). Curve 1, absorption spectrum of unbleached extract; curve 2, after 15 minutes' exposure to 390 mμ light; curve 3, after 90 minutes' exposure to 606 mμ light; curve 4, after 10 minutes' exposure to white light.

FIG. 3 B. Difference spectra of experiment with sample 11B. Upward changes indicate loss of density, downward changes gain in density. Curve 1–2 is the result of the 390 mμ bleach; curve 2–3, the 606 mμ bleach; and 3–4, the white light bleach.

mμ, indicating (pH 8.3) that retinene$_1$ is the chromophore of the pigment molecule.

The primarily marine staghorn sculpin, *Leptocottus armatus*, also has the 511$_1$ pigment. The adjusted λ_{max} obtained from consideration of the absorption spectrum was 498 to 503 mμ (Table I). Partial bleaching experiments (Fig. 3) with extracts 10 and 11, however, showed that the estimate 498 to 503 mμ is

too low. There was present in extract 11 a photostable interfering pigment, which is represented in the absorption spectrum of the unbleached extract (Fig. 3A, curve 1). Even after exposure to white light this component was still present (curve 4). A series of partial bleaching experiments, with and without NH_2OH, established both the homogeneity of the photosensitive component (Fig. 3) and that it is a 511_1 pigment. There is an extremely close fit at all wavelengths between the difference spectrum (Fig. 4, curve 3) and the Dartnall (1953) nomogram curve constructed for a visual pigment with λ_{max} 511 mμ

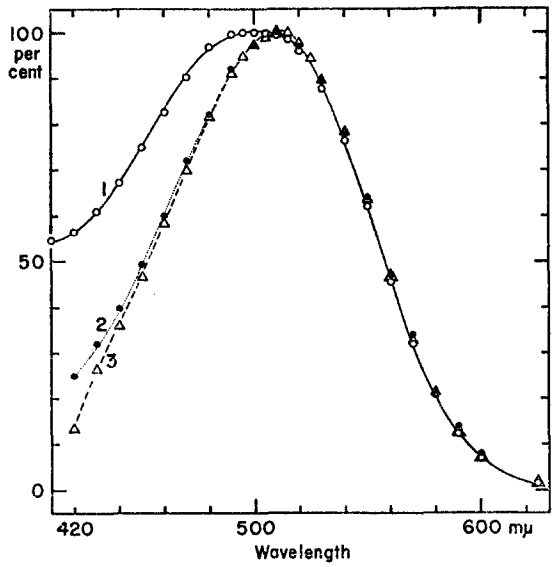

FIG. 4. Curve 1, absorption spectrum of sample 11A of *Leptocottus armatus*; curve 2, constructed from Dartnall's nomogram, assuming a maximum at 511 mμ; curve 3, NH_2OH experiment, combined difference spectrum after exposure of sample 11A to 640, 631, 606, and 580 mμ light.

(curve 2). This supports the view that 511 mμ is the true λ_{max}. The absorption spectrum (curve 1) is much too broad, owing to the presence of the stable interfering pigment. The spectral locations of both oxime (369 mμ) and product of bleaching (378 mμ) show that $retinene_1$ is the chromophore. The evidence strongly indicates, therefore, that both *Cottus* and *Leptocottus* have the same photosensitive retinal pigment.

3. Two Viviparous Perches

Retinal extracts of adult bay blackperch, *Embiotoca jacksoni*, and walleye surfperch, *Hyperprosopon argenteum*, both contained 506_1 pigments. The extracts had purity ratios of 0.65 to 0.70; adjusted λ_{max} of the absorption

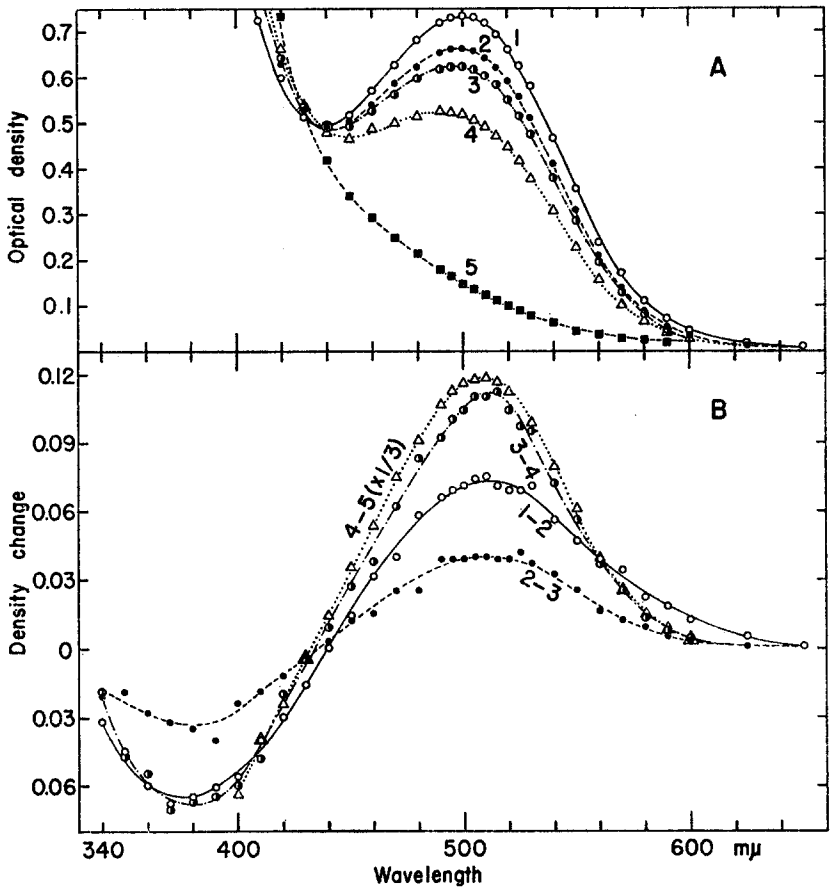

FIG. 5 A. A partial bleaching experiment with sample 7A of *Embiotoca jacksoni*. Curve 1, absorption spectrum of unbleached extract; curve 2, after 1 hours' exposure to 640 mμ light; curve 3, after 1 hours' further exposure to 640 mμ light; curve 4, after 15 minutes' exposure to 606 mμ light; curve 5, after 30 minutes' exposure to 580 mμ light.

FIG. 5 B. Difference spectra of experiment with sample 7A. Upward changes indicate loss of density, downward changes gain in density. Curves 1–2 and 2–3 are the results of the 640 mμ bleaches; 3–4, the 606 mμ bleach; and 4–5 (drawn to one-third scale), the 580 mμ bleach.

spectra were 502 to 507 mμ (Table I). Difference spectra obtained in a partial bleaching experiment with sample 7A of *Embiotoca* are substantially similar (Fig. 5), but the presence of a small proportion of a more red-sensitive pigment was suggested by the first bleach (curve 1–2). This was confirmed with sample 7B of the same extract, with which a difference spectrum maximal at 530 mμ

was obtained by exposure to deep red light. The true spectral location of this subsidiary pigment was not determined, for it constituted no more than about 5 per cent of the total and was not bleached without involvement of the chief pigment. The true λ_{max} of the less red-sensitive component was estimated from the partial bleaching experiments to be 506 mμ. The product of bleaching (378 to 380 mμ) was that of a retinene$_1$ pigment. The partial bleaching experiment with sample 8A of *Hyperprosopon* gave evidence of a 506$_1$ pigment. There

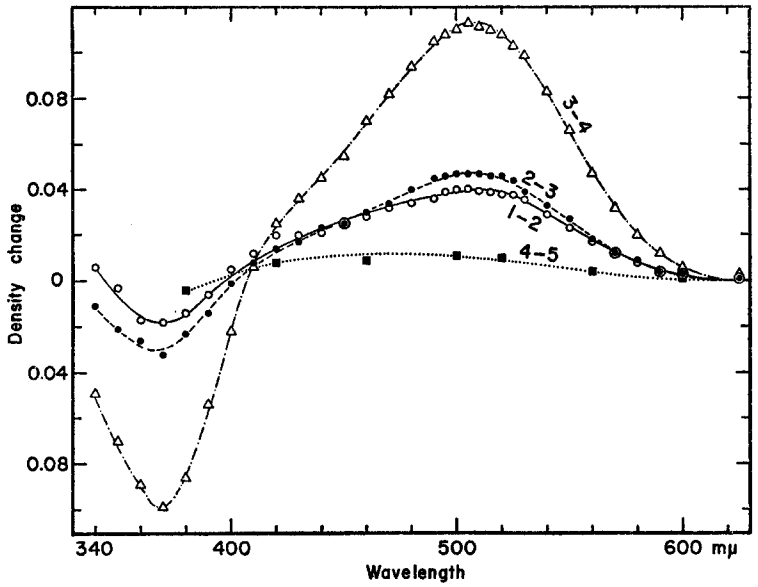

FIG. 6. Difference spectra of a partial bleaching experiment with sample 2A of *Atherinops affinis* (NH_2OH added). Upward changes indicate loss of density, downward changes gain in density. Curves 1–2 and 2–3 are the results of 50 minute and 90 minute exposures to 640 mμ light; 3–4, a 50 minute exposure to 606 mμ light; and 4–5 a final 10 minute exposure to white light.

appeared to be a small amount of a more red-sensitive pigment in this species also.

4. *Three Members of the Order Percesoces*

Retinal extracts of two atherinid members of this order were examined, and both contained homogeneous photosensitive pigments with λ_{max} 508 mμ. The extracts of both topsmelt, *Atherinops affinis*, and jacksmelt, *Atherinopsis californiensis*, contained stable light-absorbing substances and had correspondingly high purity ratios (Table I). Partial bleaching in NH_2OH experiments with samples 2A and 2B of *Atherinops* resulted in difference spectra with λ_{max} 507 to 508 mμ (Fig. 6). The oxime had λ_{max} 367 to 369 mμ, indicative of retinene$_1$. The difference spectra were very similar in a single NH_2OH experiment

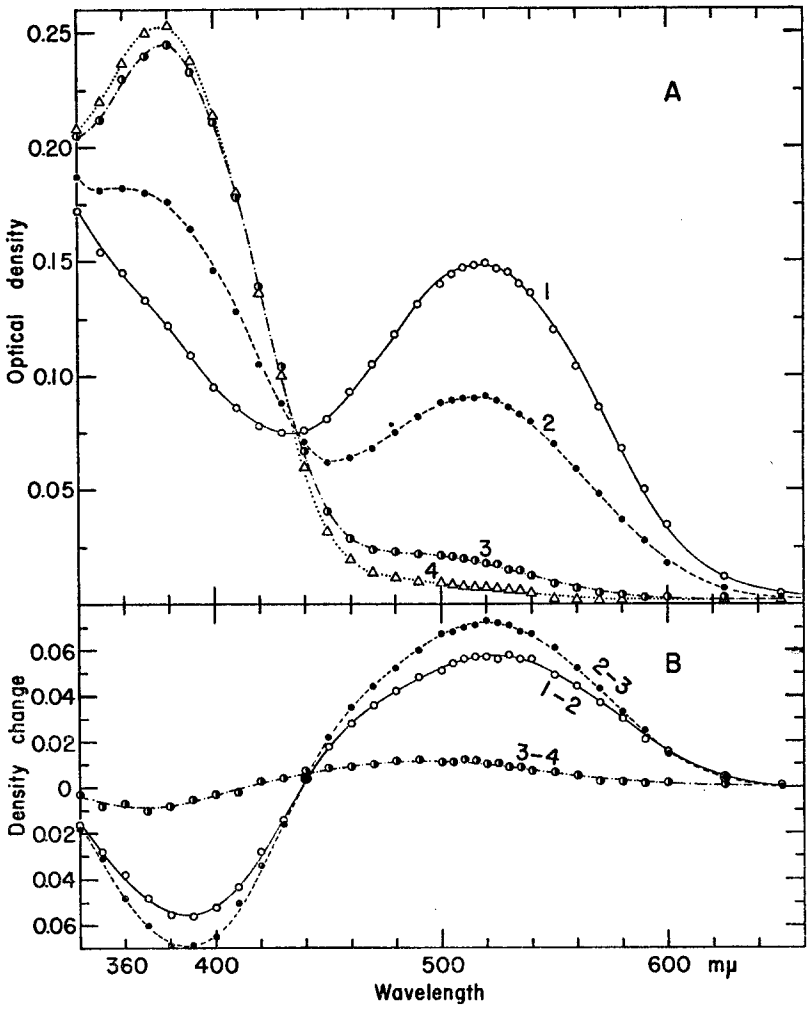

FIG. 7 A. A partial bleaching experiment with sample 4A of *Mugil cephalus* (NH_2 OH added). Curve 1, absorption spectrum of unbleached extract; curve 2, after 80 minutes' exposure to 660 mμ light; curve 3, after 40 minutes' exposure to 606 mμ light; curve 4, after 10 minutes' exposure to white light.

FIG. 7 B. Difference spectra of experiment with sample 4A. Upward changes indicate loss of density, downward changes gain in density. Curve 1–2 is the result of the 660 mμ bleach; 2–3, the 606 mμ bleach; and 3–4, the white light bleach.

with *Atherinopsis*, λ_{max} being 508 to 509 mμ. With this species the location of the oxime was unfortunately not determined, due to the presence of interfering impurities. It is therefore not known whether $retinene_1$ or $retinene_2$ is the chromophore in the retinal pigment of *Atherinopsis*.

The striped mullet, *Mugil cephalus*, is another species in the same order, but belongs to the family Mugilidae. This species, though primarily marine, has a porphyropsin with λ_{max} 520 mμ. From the absorption spectra and purity ratios of the unbleached extracts, the true λ_{max} was estimated to be 518 to 520 mμ (Table I). Partial bleaching experiments were performed with five samples of extracts 4 and 5. Both in the presence and absence of NH_2OH substantially the same results were obtained (Fig. 7). The maximum of the difference spectra

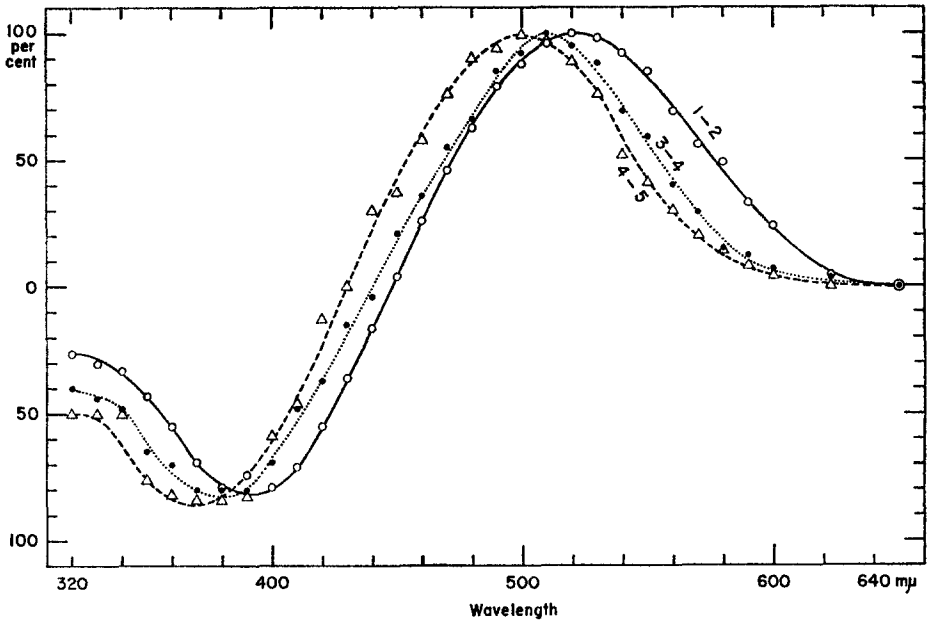

FIG. 8. A partial bleaching experiment with sample 15A of *Sphoeroides annulatus*. Difference spectra are scaled to 100 per cent for comparison. Curve 1–2, difference spectrum after 1 hour's exposure to 640 mμ light; curve 3–4, after 30 minutes' exposure to 606 mμ light; curve 4–5, after 50 minutes' further exposure to 606 mμ light. Curve 2–3, after second exposure to 640 mμ light for 1 hour, omitted for clarity of figure.

was 518 to 522 mμ, only a single component being bleached throughout the experiments. The product of bleaching absorbed light maximally at 392 to 401 mμ, the oxime at 384 to 388 mμ. These observations are similar to those reported by Crescitelli (1956) in his experiments on the bleaching products of amphibian retinene$_2$ pigments. In *Mugil* there is, therefore, a 520$_2$ retinal pigment.

5. *Mixture of Rhodopsin and Porphyropsin in a Marine Puffer*

A retinal extract of the network puffer, *Sphoeroides annulatus*, had λ_{max} 495 mμ and a purity ratio of 0.89, which produced an estimate of 508 mμ as

the true λ_{max} (Table I). This estimate, however, was not borne out by the bleaching experiment (without NH_2OH) with sample 15A. Exposure to 640 mμ light first removed a porphyropsin-like component with λ_{max} 524 mμ (Fig. 8, curve 1–2). With further exposure to 640 mμ light λ_{max} of the difference spectrum shifted to 513 mμ (curve 2–3, omitted from the figure). The sample was then exposed to 606 mμ light, and the maxima of the difference spectra shifted to 510 mμ (curve 3–4) and finally to 500 mμ (curve 4–5). There was a parallel shift in the spectral maximum of the product of bleaching from 390 to 375 mμ. This indicates that the red-sensitive component of *Sphoeroides* has retinene$_2$ as the chromophore and that the red-insensitive component has retinene$_1$. The results of this single experiment suggest that *Sphoeroides* has 500_1 and 522_2 pigments in nearly equal proportions.

6. Other Fishes from Relatively Turbid Water

Retinal extracts of two other species from sandy shore and bay habitats contained retinene$_1$ pigments with λ_{max} above 500 mμ. The California deepbody anchovy, *Anchoa compressa*, appears to have a 508_1 pigment, but the data obtained from a partial bleaching experiment with a single impure extract (1A) are not conclusive. In a retinal extract of the queenfish, *Seriphus politus*, there was a photosensitive pigment with λ_{max} 504 mμ (Table I). Homogeneity of the light-sensitive component was established in experiments with and without NH_2OH. With NH_2OH present λ_{max} of the difference spectra was 503 to 504 mμ. That the chromophore is retinene$_1$ was shown by the spectral maxima of the oxime (368 mμ) and product of bleaching (380 mμ). Both in *Anchoa* and in *Seriphus*, therefore, the photosensitive retinal pigments have a retinene$_1$ chromophore and λ_{max} above 500 mμ.

DISCUSSION

The fishes for which photosensitive retinal pigments have been described above are not similar morphologically, nor are they members of a single closely related unit. They belong to eight families placed in four orders. Within the large order Percomorphi the Sciaenidae, Embiotocidae, Cottidae, and Gobiidae represent four divergent groups. In salinity of habitat also they do not fall into a single category (see Materials and Methods). Several stages from rather strictly marine species (*Anchoa*, *Atherinopsis*, *Seriphus*, *Embiotoca*, and *Hyperprosopon*) to some that may only occasionally enter brackish water or the sea (*Cottus* and *Eucyclogobius*) are included. Yet all these species are alike in having retinal pigments with maximum absorption at wavelengths longer than 500 mμ. Thus, the results reported earlier (Munz, 1956) with *Gillichthys* are confirmed here in other members (*Clevelandia* and *Eucyclogobius*) of the same family and extended to species of a number of other groups of fishes.

Not all these species resemble *Gillichthys*, however, in having retinene$_1$ pigments with λ_{max} above 500 mμ. Although the end results in terms of spectral

absorption are similar, there are also a mixture of rhodopsin and porphyropsin in *Sphoeroides* and a porphyropsin alone in the marine but euryhaline *Mugil*. The results obtained with the different species (including *Gillichthys*) are summarized as follows:—

Seriphus politus	504_1	*Leptocottus armatus*	511_1
Embiotoca jacksoni	506_1	*Clevelandia ios*	512_1
Hyperprosopon argenteum	506_1	*Eucyclogobius newberryi*	512_1
Anchoa compressa	508_1	*Gillichthys mirabilis*	512_1
Atherinops affinis	508_1	*Sphoeroides annulatus*	$500_1 + 522_2$
Cottus sp.	511_1	*Mugil cephalus*	520_2

(*Atherinopsis californiensis* has a $508_?$ pigment.)

What then do these fishes have in common beside the general nature of their retinal pigments? They all occur in shallow, relatively turbid coastal waters that are alike in having transmission maxima at wavelengths above 500 mμ. While no measurements of Pacific coast water samples are available, the transmission maxima in waters of this type are probably similar to the 520 to 570 mμ range determined along the Atlantic coast by Clarke and James (1939) and Hulburt (1945). There seems to be a general trend for the spectral locations of the retinal pigments of fishes living in these waters to parallel the transmission maxima of the environment. It is true that *Embiotoca*, *Atherinops*, and *Atherinopsis* also live in clearer waters near rocky areas or away from shore. The specimens of *Embiotoca* and *Atherinops* examined in this study, however, were members of bay populations; *Atherinopsis* was seined on a sandy beach.

It is suggested that the retinal pigments of turbid water fishes may have become adapted to the spectral quality of light available in their habitats. Vision is an important sensory modality in almost all the species that have been examined. It seems reasonable that natural selection should operate to produce visual pigments well suited for the absorption of light available in these habitats. Even though the shift in λ_{max} is in most instances rather small, this might have a disproportionately large effect at longer wavelengths. The retinal pigments of turbid water gobies, *Clevelandia* and *Eucyclogobius* (and *Gillichthys*), may be compared with that of another goby, *Coryphopterus nicholsii*, from clearer water (Fig. 1). Even though the difference in λ_{max} is only 12 mμ, the retinal pigments of *Clevelandia* and *Eucyclogobius* are probably about twice as effective as that of *Coryphopterus* in absorbing 570 mμ light. This wavelength is perhaps that which is transmitted maximally in the habitats of *Clevelandia* and *Eucyclogobius*.

While it is not claimed that this general hypothesis is established, it is strengthened by the results obtained with another group of fishes. In certain deep sea fishes the retinal pigments are based on retinene_1, but have λ_{max} 478 to 490 mμ (Denton and Warren, 1956, 1957; Munz, 1957, 1958; see also Wald,

Brown, and Brown, 1957). This spectral shift toward shorter wavelengths parallels the predominance of blue light in the environment of bathypelagic fishes. Denton and Warren, and Munz have suggested that the retinal pigments of bathypelagic fishes may be adapted to the spectral composition of light available at moderate depths. This offers a biological interpretation of the confusing array of photosensitive retinal pigments that have been described for marine and euryhaline fishes.

Grateful acknowledgment is made to Dr. F. Crescitelli and Dr. B. W. Walker for their assistance and advice during this investigation. Much of the equipment used was made available by Dr. Crescitelli's research grant from United States Public Health Service.

REFERENCES

Clarke, G. L., and James, H. R., Laboratory analysis of the selective absorption of light by sea water, *J. Opt. Soc. America*, 1939, **29**, 43.

Crescitelli, F., The nature of the gecko visual pigment, *J. Gen. Physiol.*, 1956, **40**, 217.

Crescitelli, F., and Dartnall, H. J. A., A photosensitive pigment of the carp retina, *J. Physiol.*, 1954, **125**, 607.

Dartnall, H. J. A., Visual pigment 467, a photosensitive pigment present in tench retinae, *J. Physiol.*, 1952, **116**, 257.

Dartnall, H. J. A., The interpretation of spectral sensitivity curves, *Brit. Med. Bull.*, 1953, **9**, 24.

Denton, E. J., and Warren, F. J., Visual pigments of deep-sea fish, *Nature*, 1956, **178**, 1059.

Denton, E. J., and Warren, F. J., The photosensitive pigments in the retinae of deep-sea fishes, *J. Marine Biol. Assn. United Kingdom*, 1957, **36**, 651.

Hulburt, E. O., Optics of distilled and natural water, *J. Opt. Soc. America*, 1945, **35**, 698.

Jones, I. C., and Eckstein, P., editors, The Comparative Endocrinology of Vertebrates. Part II. The Hormonal Control of Water and Salt-Electrolyte Metabolism in Vertebrates, Cambridge at the University Press, 1956.

Morton, R. A., and Pitt, G. A. J., The visual pigments, *Progr. Chem. Org. Nat. Products*, 1957, **14**, 244.

Munz, F. W., A new photosensitive pigment of the euryhaline teleost, *Gillichthys mirabilis*, *J. Gen. Physiol.*, 1956, **40**, 233.

Munz, F. W., Photosensitive pigments from retinas of deep-sea fishes, *Science*, 1957, **125**, 1142.

Munz, F. W., Photosensitive pigments from the retinae of certain deep-sea fishes, *J. Physiol.*, 1958, **140**, 220.

Wald, G., The biochemistry of vision, *Ann. Rev. Biochem.*, 1953, **22**, 497.

Wald, G., Brown, P. K., and Brown, P. S., Visual pigments and the depths of habitat of marine fishes, *Nature*, 1957, **180**, 969.